The Effect of Water-Extracted Solubles from Gluten on Its Baking and Rheological Properties[1]

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ABSTRACT

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Rheologically active material can be removed from commercial dry gluten by washing it with distilled water. Control and washed glutens performed equally in baking tests when they were added to flour. However, the control (unwashed) gluten performed better when baked as gluten/starch doughs. When only 1.3% of the gluten was removed, dynamic rheological tests showed that the washed gluten had higher G' (more resistant to deformation) and lower loss tangent values (G''/G', relatively more elastic and less viscous) than the control gluten. Mixograph and dynamic rheological tests showed that the washed gluten gave no response to mixing with iodate, whereas the control gluten had an iodate response. This suggests that the soluble material removed from the washed gluten is involved in the effect of KIO_3. The nature of the material is unknown. Two gluten washing methods (centrifugation to develop a dough, and mixing) were used to prepare glutens from seven different flours. For all these flours, glutens prepared by centrifugation had longer mix times than glutens prepared by mixing. Glutens washed from soft wheat flours by centrifugation did not form cohesive doughs. Glutens prepared from hard wheat flours by centrifugation formed cohesive doughs, and the dynamic rheological properties of these doughs were all essentially the same. All glutens produced by the mixing method formed cohesive doughs, and there were significant differences in G' between those glutens produced from different flours. Thus, the rheological properties of glutens were dependent upon the method of preparation.

Dreese et al (1988a) reported that soluble materials could be removed from commercial gluten by washing with distilled water. They further reported that the washed commercial gluten produce doughs with higher G' values and lower loss tangent values than doughs made from the control gluten (i.e., the washed gluten doughs are more resistant to deformation and are relatively more elastic and less viscous). Washing ratios (distilled water/gluten) of 3:1 and 10:1 were used. The 10:1 washing ratio caused a much larger rheological effect than did the 3:1 washing ratio. The 3:1 and 10:1 washing ratios removed 2.0 and 2.9% solubles, respectively, from the commercial gluten. These findings suggested that a two-stage washing procedure might concentrate a rheologically active component in the solubles. Dreese et al (1988a) did not examine the solubles washed from the commercial gluten or the effect of washing on the gluten's baking properties. Because removal of only 2.9% solubles had a rather large rheological effect, it seemed reasonable that understanding the solubles might be helpful in understanding gluten quality.

Schroeder and Hoseney (1978) used a mixograph to study the effect of potassium iodate (KIO_3) on wheat flour doughs. They reported that iodate had no effect on gluten-starch doughs. Their method of gluten separation involved slurrying the flour with water (10:1, water/flour) and then centrifuging.

In this study, we examined properties of the soluble material washed from commercial gluten and the effect of washing on gluten's baking properties. We also determined the effect of flour source and flour washing method on the rheological properties and response to oxidation with KIO_3 of handwashed lyophilized gluten.

MATERIALS AND METHODS

Materials

The commercial gluten used was from Midwest Grain Products (Atchison, KS). Dry basis protein and ash contents were 81.0 and 1.0%, respectively.

Gluten was washed from the seven flours described in Table I. Flours A, B, C, and D were commercial straight-grade flours. Flours K1, K2, and K3 were milled on the Kansas State University pilot mill to approximately 72% extraction.

All water used was distilled.

Washing of Commercial Gluten

Gluten (500 or 150 g for 3:1 or 10:1 washing ratios, respectively) and 1,500 ml of water were placed in a 4-L jar with a tight-fitting lid. The jar and contents were shaken vigorously by hand for 2 min and then centrifuged for 15 min at 1,500 × *g*. The supernatant and centrifugate each were lyophilized to produce the solubles and washed gluten from the 3:1 or 10:1 washing procedures. A two-stage washing procedure also was used. The centrifugate from the 3:1 wash was divided into three equally sized pieces. One piece was placed in a blender (Osterizer) along with 500 ml of water. The blender was run for 15 sec at high speed, and the resulting slurry was poured into a 4-L jar. The procedure was repeated for the other two gluten pieces, and then all three slurries were combined in the same jar. The jar was sealed and shaken vigorously for 2 min. The resulting slurry was centrifuged for 15 min at 1,500 × *g*. The supernatant and centrifugate were lyophilized to give what will be called 3+10 solubles and 3+10 washed gluten, respectively. After lyophilization, all washed glutens were ground first in a blender (Osterizer) and then in a Udy cyclone mill with a 1-mm screen.

Washing Gluten from Flour by Centrifuging

Five hundred grams of flour and 1,500 ml of water were placed in a 4-L jar, sealed, and shaken vigorously by hand for 2 min. The resulting slurry was centrifuged for 15 min at 1,500 × *g*. The centrifugate was massaged by hand in 1,500 ml of water. Hard wheat glutens held together in a cohesive mass, and the wash water and dispersed starch were poured off. Soft wheat glutens did not form a cohesive mass and were separated from the wash water and dispersed starch by sieving through a U.S. no. 30 (600 μm) sieve. This washing process was repeated twice. The three washing processes used 12 L of water per kilogram of flour.

TABLE I
Flour Description and Analytical Values

Flour	Class[a]	Protein[b]	Ash[b]	Moisture
A	HRS	13.1	0.54	11.4
B	HRW	11.0	0.46	12.7
C	SRW	8.4	0.45	12.9
D	HRW	8.8	0.42	13.0
K1	HRW	9.8	0.46	12.1
K2	HRW	12.1	0.46	12.9
K3	HRW	10.7	0.42	12.9

[a] HRW = Hard red wheat; SRW = soft red wheat.
[b] Values on a 14% moisture basis.

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Washing Gluten from Flour by Mixing

Flour (150 g) and 90 ml of water were mixed to optimum development in a pin mixer (TMCO National Manufacturing Lincoln, NE). Three of these doughs were combined and placed in 900 ml of water. Doughs were massaged by hand as with the centrifuge method. Hard and soft wheat glutens all formed a cohesive mass, and the wash water and dispersed starch were poured off. The three washing processes used 6.6 L of water per kilogram of flour washed.

Glutens from the centrifugation and the mixing methods were lyophilized, ground in a blender (Osterizer), and then ground on a Udy mill with a 1.0-mm screen.

Rheological Testing

Doughs were tested in a dynamic rheometer as described by Faubion et al (1985).

Baking Tests

Baking tests were conducted using the pup loaf procedure described by Finney (1984) with a 180 min fermentation time. The control formula contained 100 g (14% mb) of flour, 0.76 g of instant dry yeast (Fermipan), 6 g of sugar, 1.5 g of salt, 3 g of shortening, 4 g of nonfat dry milk, optimum potassium bromate, and optimum water. Gluten was evaluated by adding 3 g of gluten to the control formula and by replacing flour in the control formula with 81 g of commercial unmodified wheat starch (Midwest Grain Products Atchison, KS) and 19 g of gluten. Water and bromate were adjusted to optimum for all formula variations. Flour B (Table I) was used for baking tests.

Statistical Analysis

All rheological values reported are the means of at least two, and generally more, replicate tests. Standard deviations between replicate doughs were 0.021 log units for G' and 0.023 units for the loss tangent. Loaf volumes reported are means of at least three loaves. Standard deviations between replicate loaves were 10 cm^3 for loaf volume measurements.

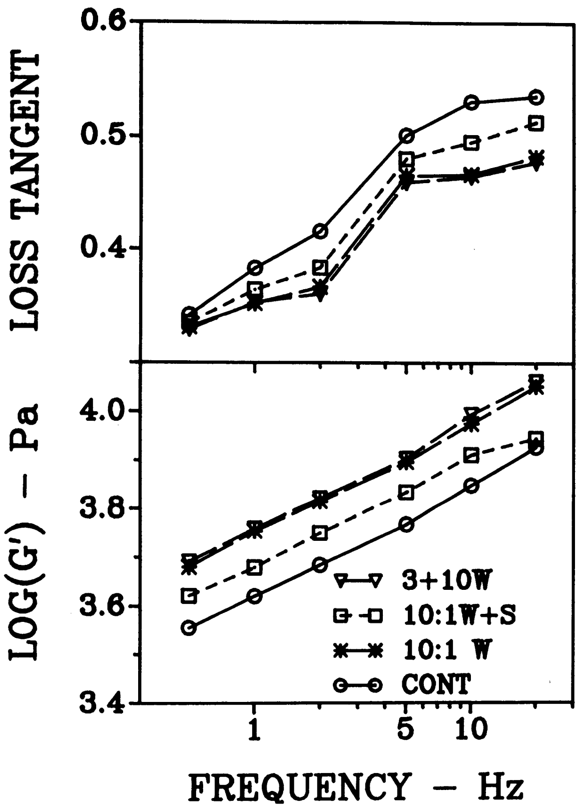

Fig. 1. Loss tangent and G' for gluten/water doughs at 57% moisture. CONT = control, 10:1 W = gluten washed 10:1 as explained in text, 10:1W+S = 10:1 washed gluten with solubles added back, 3+10W = gluten washed 3+10 as explained in text.

RESULTS AND DISCUSSION

Stability and Rheological Effect of Solubles

The solubles from the 10:1 wash of gluten always collapsed and turned slightly brown during freeze-drying, whereas the 3:1 solubles did not collapse and had a white color when dried. Adding the 10:1 solubles back to the 10:1 washed gluten caused a decrease in G' (Fig. 1) but did not return G' to the level of the unwashed gluten control. It is possible that the 10:1 solubles suffered a partial loss of activity because of the collapse during freeze-drying.

The 3+10 washing and drying procedure was replicated several times. The 3+10 solubles usually remained glassy during lyophilizing and dried to a white color. Occasionally the 3+10 solubles collapsed during lyophilizing and had a brownish color when dried. As a precaution, with all tests reported here, only white 3+10 solubles were used. The dry weight of the 3+10 solubles was 1.3 g for each 100 g (14% moisture basis) of original gluten washed.

Rheological tests showed the 3+10 washed gluten not to be significantly different from the 10:1 washed gluten (Fig. 1). When the 3+10 solubles were added to the 3+10 washed gluten, results were dependent on the freshness of the solubles. If the solubles were tested within hours after removal from the lyophilizer, G'

TABLE II
Baking Results

Sample[a]	Mix Time (min)	$KBrO_3$ (ppm)	Absorption (%)	Loaf Volume (cm^3)
Control (100% flour)	4.3	20	62.5	960
100% Flour + 3% CG	4.3	20	65.0	1,024
100% Flour + 3% WCG	4.3	20	65.0	1,022
81% Starch + 19% CG	6.0	0	62.5	675
81% Starch + 19% WCG	12.0	0	62.5	565

[a] Flour is B in Table I. CG is commercial gluten. WCG is 10:1 washed commercial gluten.

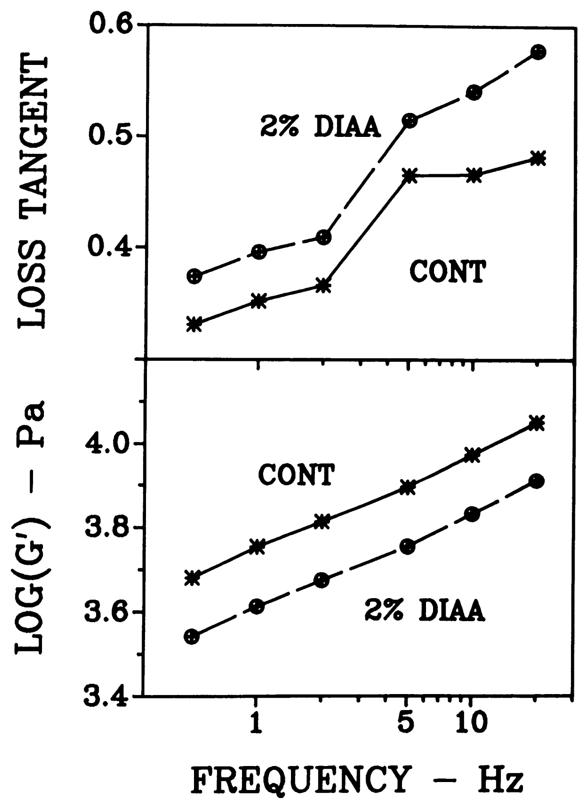

Fig. 2. Loss tangent and G' for gluten/water doughs at 57% moisture. CONT = control, 2% DIAA dough = 2% gluten basis of D-isoascorbic acid.

was reduced to essentially that of the control. However, after the solubles were stored at 3°C for two weeks or longer, they produced no rheological effect.

D-Isoascorbic Acid

We speculated that the active component in the 3+10 solubles was oxidized by the air, causing the loss of activity during storage. D-isoascorbic acid (DIAA) is oxidizable and acts as a reducing agent in dough (Lillard et al 1982). The 2% DIAA added to doughs made from 10:1 washed gluten and water caused a decrease in *G′* and an increase in tangent (Fig. 2) similar in magnitude to that caused by 1.3% of the 3+10 solubles. Therefore, the active ingredient in the 3+10 solubles is more effective than the DIAA even if we assume that 1.3% is all the active material, although this appears highly unlikely.

TABLE III
Amino Acid Composition of 3+10 of Solubles

Amino Acid	Grams per 100 g of Protein
Aspartic acid	4.0
Threonine	2.4
Serine	4.9
Glutamic acid	28.1
Proline	12.4
Glycine	2.8
Alanine	3.1
Half cystine	2.7
Valine	3.3
Methione	3.0
Isoleucine	2.9
Leucine	5.8
Tyrosine	4.1
Phenylalanine	5.8
Histidine	3.5
Lysine	1.7
Ammonia	3.8
Arginine	5.8

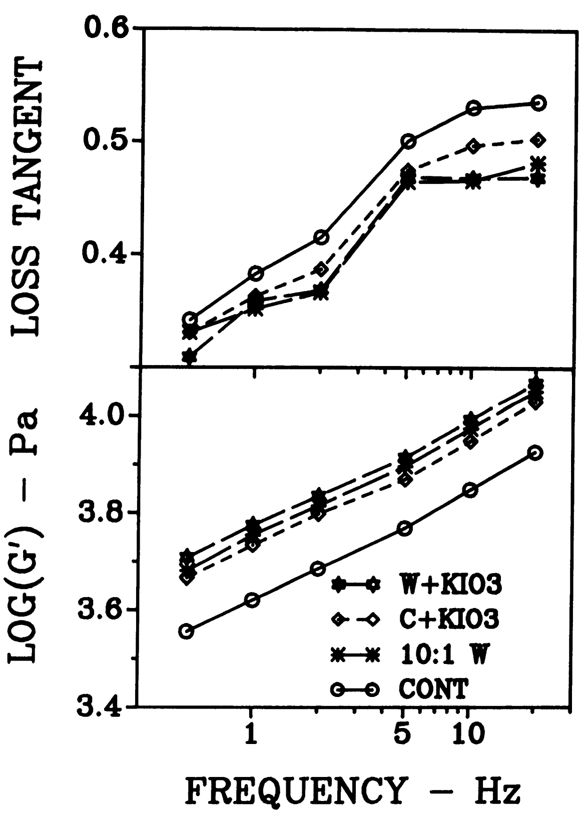

Fig. 3. Loss tangent and *G′* for gluten/water doughs at 57% moisture. CONT = control, 10:1W = 10:1 washed gluten as explained in text, C+KIO_3 = control gluten with 50 ppm gluten basis of potassium iodate, W+KIO_3 = 10:1 washed gluten with 50 ppm potassium iodate.

Baking Tests with Washed Gluten

Results (Table II) showed that when added to flour, the 10:1 washed commercial gluten (WCG) and control commercial gluten (CG) performed similarly. When CG was added to starch, however, it produced bread with higher volume and better grain than starch with WCG. The CG-starch bread had a slight overoxidized appearance, whereas the WCG-starch bread was strongly overoxidized. This is in agreement with the rheological observation that the solubles washed from the gluten have a rheological effect similar to that of cysteine (Dreese et al 1988b).

Flour must contain the compounds that are washed from commercial gluten. It is possible that these compounds have an effect only up to a certain level. This would explain why the CG and WCG performed equally when added to flour but CG performed better when added to starch.

Composition of 3+10 Solubles

The data in Figure 1 showed 3+10 solubles to be potent rheologically. An amino acid analysis indicated the 3+10 solubles contained 53.1% protein with the amino acid composition shown in Table III. The glutamic acid level (28.1%) is somewhat lower than that expected for gluten. The low level of soluble material (1.3%), of which only half is protein, suggests that a component other than protein might be responsible for the rheological effect.

Effect of Iodate

Schroeder and Hoseney (1978) showed that when flour is extracted with an excess of water (1:10), the flour no longer breaks down rapidly (thin mixogram tail) when mixed with KIO_3. Therefore, to determine if the water-soluble factor from flour was the same factor as in gluten, water doughs were made with commercial gluten and 10:1 washed commercial gluten. Doughs from each were made without KIO_3 or with 50 ppm (gluten basis) KIO_3. Addition of KIO_3 had no significant effect on the 10:1 washed commercial gluten but caused an increase in *G′* and a decrease in the loss tangent of the commercial gluten doughs (Fig. 3). Therefore, these data indicate that the solubles removed from the CG by washing were involved in the mechanism of the iodate effect.

Mixograph Study of Iodate

Mixograph tests of gluten/starch (19:81) doughs were conducted with and without 50 ppm (gluten + starch basis) KIO_3. Because the gluten/starch doughs had long development and breakdown times, the mixograph was run for 25 min. Development times were shorter, and the mixograph "tail" was thinner for the control gluten doughs than for the 10:1 washed gluten doughs (Fig. 4). Iodate had no effect on the 10:1 washed gluten doughs, in agreement with the report of Schroeder and Hoseney (1978). However, iodate did cause the tail of the control (unwashed) gluten dough to become much thinner than that of the control gluten dough without iodate.

Comparison of Flour/Water Doughs

It is generally believed by gluten producers that good quality gluten can be produced from essentially any flour. Therefore a number of flours and two gluten washing methods were investigated. The flours were characterized by mixograms (Fig. 5), which reflect differences in the protein quantity and quality among the flours.

Dreese et al (1988b) reported that dynamic rheological tests of flour/water doughs do not show differences between a hard wheat and a soft wheat flour when studied at the same dough moisture content. Their study used only one hard wheat and one soft wheat flour. In this study, five hard and two soft wheat flours were tested. The two soft wheat flours had lower *G′* values than any of the hard wheat flours tested. This may only reflect their lower protein contents.

Handwashed Lyophilized Glutens

Protein contents and mixing times for the handwashed lyophilized glutens are shown in Table IV. The glutens produced from soft wheat by the centrifuge method were low in protein. The amount of wash water used was the same for hard and soft wheat flours, and the amount of hand agitation was held as constant as possible. The soft wheat glutens tended to disintegrate during washing. Another possible explanation for the low protein content of the centrifuged soft wheat glutens was that soft wheat starch and gluten do not separate easily if they have not been previously mixed into a dough.

TABLE IV
Descriptions of Handwashed Lyophilized Glutens

Wash Method/ Flour[a]	Protein[b]	Mix Time[c]
Centrifuged		
A	78.7	9.5
B	77.1	8.0
C	43.2	...[d]
D	42.1	...[d]
K1	74.8	9.5
K2	69.5	9.0
K3	69.5	9.5
Mixed		
A	78.1	1.3
B	83.4	1.5
C	74.5	1.2
D	79.7	1.6
K1	88.9	1.2
K2	81.4	1.5
K3	89.4	1.4

[a] Flour descriptions as in Table I.
[b] Dry basis.
[c] Mixing was in a pin mixer (TMCO National Manufacturing, Lincoln, NE). Optimum mix time was subjectively judged.
[d] These glutens were mixed 30 min but did not form a cohesive dough.

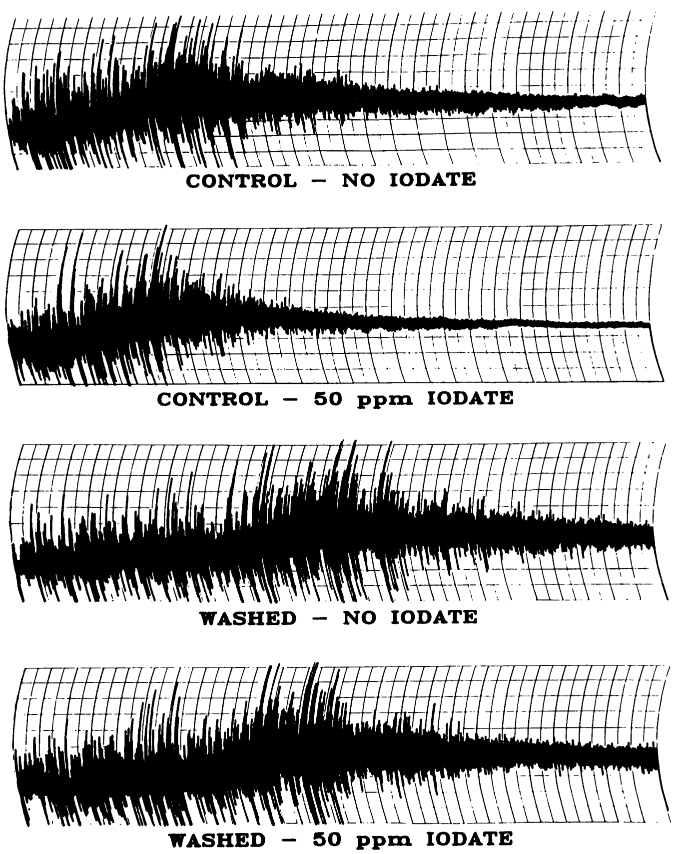

Fig. 4. Mixographs of gluten/starch (19:81) doughs at 60% water absorption. CONTROL = unwashed commercial gluten. WASHED = 10:1 washed gluten as indicated in text. Potassium iodate added as indicated.

Mix times to produce a gluten-starch dough were consistently less for the glutens prepared by the mixing method than for the gluten prepared by the centrifuge method. Mixing-time differences among glutens from different flours were minor if the same washing procedure was used. An exception was the soft wheat glutens washed by the centrifuge procedure, which did not mix to good development in 30 min.

Dynamic rheological tests showed no significant differences between centrifuged glutens washed from the five hard wheat flours. The centrifuged soft wheat glutens would not form a cohesive dough and, therefore, were not tested.

Dynamic rheological tests of the glutens prepared by the mixing procedure showed differences (Fig. 6). The glutens from soft wheat flour had lower G' values than did similarly prepared glutens from hard wheat flours. G' values for the hard red spring gluten (A) were lower than for the hard red winter glutens (B, K2, K3). There were no significant differences in G' values within wheat class or in loss tangent values of any of the glutens tested.

Effect of Potassium Iodate on Centrifuged or Mixed Gluten

It was shown previously (Fig. 4) that potassium iodate altered the rheological properties of doughs made from commercial gluten but did not affect doughs made from 10:1 washed commercial gluten. The glutens prepared by the centrifuge and mixing methods from flour B were tested both with and without iodate. Rheometer

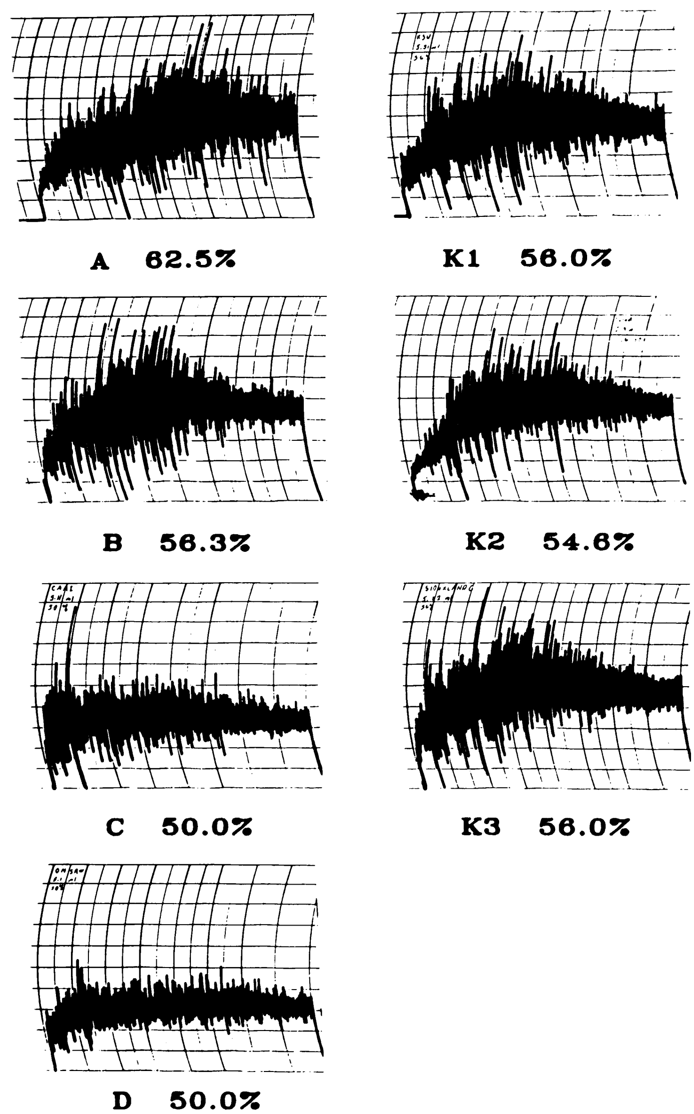

Fig. 5. Mixographs of flours described in Table I. Absorptions are as indicated.

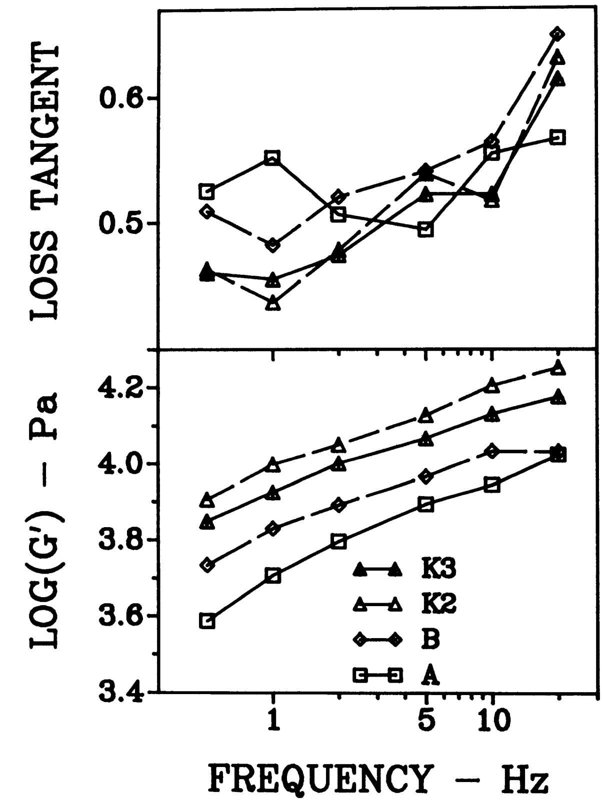

Fig. 6. Loss tangent and G' for gluten/water doughs at 57% moisture. Glutens were washed from flour by the mixing method as explained in text. Source flours are described in Table I.

tests showed that iodate caused an increase in G' for doughs prepared from glutens washed by the mixing procedure but did not affect doughs prepared from glutens washed by the centrifuge procedure. Iodate did not affect the loss tangent values for any of the doughs.

CONCLUSIONS

A material is extractable in excess water from gluten that affects the rheology of gluten. In addition, this material appears to be related to the effect of KIO_3 in doughs. The nature of the material is unknown.

Gluten was difficult to wash from soft wheat flour unless it had been first mixed to a dough. The rheological properties of glutens were dependent on the methods of preparation.

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